

#3

SEQUENCE LISTING

<110> Rosenblum et al.  
<120> Immunotoxins Directed Against c-erbB-2 (HER-2/Neu)  
Related Surface Antigens  
<130> D5425CIP2  
<140>  
<141> 1999-05-26  
<150> 08/404,499  
<151> 1995-03-17  
<160> 14  
  
<210> 1  
<211> 5  
<212> PRT  
<213> Artificial Sequence  
<220>  
<223> synthetic linker sequence  
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Gly Gly Gly Gly Ser  
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<210> 2  
<211> 18  
<212> DNA  
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<223> Primer directed towards 5' upstream region of TAb 250  
heavy chain  
<400> 2  
atatagcagg accatatg 18  
  
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<220>

<223> Primer directed towards coding region of TAb 250 heavy chain

<400> 3

atgaacttgg ggctc 15

<210> 4

<211> 15

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer directed towards 5' upstream region of TAb 250 light chain

<400> 4

tttacttcct tattt 15

<210> 5

<211> 15

<212> DNA

<213> Artificial Sequence

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<223> Primer directed towards 5' coding region of TAb 250 light chain

<400> 5

atgggcatca agatg 15

<210> 6

<211> 33

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer directed towards sFv-23

<400> 6

gctgcccaac cagccatggc gatgtctgac gtc 33

<210> 7

<211> 37  
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<210> 8  
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 <223> Primer directed towards gelonin  
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<210> 9  
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 <223> Primer directed towards gelonin  
 <400> 9  
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<210> 10  
 <211> 1517  
 <212> DNA  
 <213> Artificial  
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 <223> nucleotide sequence encoding scFv23-gelonin  
 immunotoxin  
 <400> 10  
 cccatggcga tgtctgacgt ccagctgacc cagtctccag caatcctgtc tgcattctcca 60  
 ggggagaagg tcacaatgac ttgcagggcc accccaagtg taagttacat gcactggtat 120

cagcagaagc	caggatcctc	ccccaacct	tggatttata	ccacatccaa	cctgggttct	180
ggagtccttg	ctcgcttcag	tggcgggtggg	tctgggacct	cttactctct	cacagcagca	240
gagtggaggc	tgaagatgct	gccacttatt	actgccagca	gtggagtcgt	agcccaccca	300
cgttcggagg	ggggtccaag	ctggaaataa	aaggttctac	ctctggttct	ggtaaattct	360
ctgaaggtaa	aggtgtgcag	ctgcaggagt	caggacctga	ggtggtgaag	cctggagggt	420
caatgaagat	atcctgcaag	acttctgggt	actcattcac	tggccacacc	atgaactggg	480
tgaagcagag	ccatggaaag	aaccttgagt	ggattggact	tattaatcct	tacaatggtg	540
atactaacta	caaccagaag	ttcaagggca	aggccacatt	tactgtagac	aagtcgtcca	600
gcacagccta	catggagctc	ctcagtctga	catctgagga	ctctgcagtc	tattactgtg	660
caaggagggg	tacggactgg	tacttcgatg	tctggggcgc	agggaccacg	gtcaccgtct	720
cctcagctag	cgggtggcgt	ggctccggtc	tagataccgt	tagcttcagc	accaaaggcg	780
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gtaacagcca	tggcatcccc	ctgctgcgta	aaggtgatga	cccgggtaaa	tgcttcgtgc	900
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cagcgtacga	aggcctgttc	aaaaacacca	tcaaaaaccc	gctgctgttc	ggtggcaaaa	1080
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tcgacaacta	caaaccgacc	gaaatcgcg	gctctctggg	tgtgatccag	atgggtgagcg	1260
aagcggcacg	tttcaccttc	atcgaaaacc	agattcgtaa	caacttccag	cagcgtatcc	1320
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gtaccagcgg	tgcgaacggg	atgttcagcg	aagcggtgga	actggaacgc	gcgaacggca	1440
aaaaataacta	cgtgactgcg	gtggatcagg	tgaaaccgaa	aatcgcactg	ctgaaattcc	1500
tcgagaaaaga	cgagctg					1517

<210> 11

&lt;212&gt; PRT

$\langle 220 \rangle$

<400> 11

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Leu	Ser	Ala	Ser	Pro	Gly	Glu	Lys	Val	Thr	Met	Thr	Cys	Arg	Ala
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Thr	Pro	Ser	Val	Ser	Tyr	Met	His	Trp	Tyr	Gln	Gln	Lys	Pro	Gly
				35					40					45
Ser	Ser	Pro	Lys	Pro	Trp	Ile	Tyr	Thr	Thr	Ser	Asn	Leu	Ala	Ser
				50					55					60
Gly	Val	Pro	Ala	Arg	Phe	Ser	Gly	Gly	Gly	Ser	Gly	Thr	Ser	Tyr
				65					70					75
Ser	Leu	Thr	Val	Ser	Arg	Val	Glu	Ala	Glu	Asp	Ala	Ala	Thr	Tyr
				80					85					90
Tyr	Cys	Gln	Gln	Trp	Ser	Arg	Ser	Pro	Pro	Thr	Phe	Gly	Gly	Gly
				95					100					105
Ser	Lys	Leu	Glu	Ile	Lys	Gly	Ser	Thr	Ser	Gly	Ser	Gly	Lys	Ser
				110					115					120
Ser	Glu	Gly	Lys	Gly	Val	Gln	Leu	Gln	Glu	Ser	Gly	Pro	Glu	Val
				125					130					135
Val	Lys	Pro	Gly	Gly	Ser	Met	Lys	Ile	Ser	Cys	Lys	Thr	Ser	Gly
				140					145					150
Tyr	Ser	Phe	Thr	Gly	His	Thr	Met	Asn	Trp	Val	Lys	Gln	Ser	His
				155					160					165
Gly	Lys	Asn	Leu	Glu	Trp	Ile	Gly	Leu	Ile	Asn	Pro	Tyr	Asn	Gly
				170					175					180
Asp	Thr	Asn	Tyr	Asn	Gln	Lys	Phe	Lys	Gly	Lys	Ala	Thr	Phe	Thr
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Val	Asp	Lys	Ser	Ser	Ser	Thr	Ala	Tyr	Met	Glu	Leu	Leu	Ser	Leu			
				200					205					210			
Thr	Ser	Glu	Asp	Ser	Ala	Val	Tyr	Tyr	Cys	Ala	Arg	Arg	Val	Thr			
				215					220					225			
Asp	Trp	Tyr	Phe	Asp	Val	Trp	Gly	Ala	Gly	Thr	Thr	Val	Thr	Val			
				230					235					240			
Ser	Ser	Ala	Ser	Gly	Gly	Gly	Gly	Ser	Gly	Leu	Asp	Thr	Val	Ser			
				245					250					255			
Phe	Ser	Thr	Lys	Gly	Ala	Thr	Tyr	Ile	Thr	Tyr	Val	Asn	Phe	Leu			
				260					265					270			
Asn	Glu	Leu	Arg	Val	Lys	Leu	Lys	Pro	Glu	Gly	Asn	Ser	His	Gly			
				275					280					285			
Ile	Pro	Leu	Leu	Arg	Lys	Gly	Asp	Asp	Pro	Gly	Lys	Cys	Phe	Val			
				290					295					300			
Leu	Val	Ala	Leu	Ser	Asn	Asp	Asn	Gly	Gln	Leu	Ala	Glu	Ile	Ala			
				305					310					315			
Ile	Asp	Val	Thr	Ser	Val	Tyr	Val	Val	Gly	Tyr	Gln	Val	Arg	Asn			
				320					325					330			
Arg	Ser	Tyr	Phe	Phe	Lys	Asp	Ala	Pro	Asp	Ala	Ala	Tyr	Glu	Gly			
				335					340					345			
Leu	Phe	Lys	Asn	Thr	Ile	Lys	Asn	Pro	Leu	Leu	Phe	Gly	Gly	Lys			
				350					355					360			
Thr	Arg	Leu	His	Phe	Gly	Gly	Ser	Tyr	Pro	Ser	Leu	Glu	Gly	Glu			
				365					370					375			

Lys Ala Tyr Arg Glu Thr Thr Asp Leu Gly Ile Glu Pro Leu Arg  
 380 385 390

Ile Gly Ile Lys Lys Leu Asp Glu Asn Ala Ile Asp Asn Tyr Lys  
 395 400 405

Pro Thr Glu Ile Ala Ser Ser Leu Val Val Ile Gln Met Val Ser  
 410 415 420

Glu Ala Ala Arg Phe Thr Phe Ile Glu Asn Gln Ile Arg Asn Asn  
 425 430 435

Phe Gln Gln Arg Ile Arg Pro Ala Asn Asn Asn Ile Ser Leu Glu  
 440 445 450

Asn Lys Trp Gly Lys Leu Ser Phe Gln Ile Arg Thr Ser Gly Ala  
 455 460 465

Asn Gly Met Phe Ser Glu Ala Val Glu Leu Glu Arg Ala Asn Gly  
 470 475 480

Lys Lys Tyr Tyr Val Thr Ala Val Asp Gln Val Lys Pro Lys Ile  
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Ala Leu Leu Lys Phe Leu Glu Lys Asp Glu Leu  
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<210> 12

<211> 1500

<212> DNA

<213> Artificial

<220>

<223> nucleotide sequence encoding scFv23-gelonin  
 immunotoxin

<400> 12

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gtcacaatga	cttgcagggc	cacccaagt	gtaagttaca	tgcactggta	tcagcagaag	120
ccaggatcct	cccccaaacc	ttggatttat	accacatcca	acctggcttc	tggagtcctt	180
gctcgcttca	gtggcggtgg	gtctggggacc	tcttactctc	tcacagtcag	cagagtgagg	240
gctgaagatg	ctgccactta	ttactgccag	cagtggagtc	gtagcccacc	cacgttcgga	300
gggggggtcca	agctggaaat	aaaagggttct	acctctgggt	ctggtaaata	ttctgaagggt	360
aaagggtgtgc	agctgcagga	gtcaggacct	gaggtgggtga	agcctggagg	ttcaatgaag	420
atatcctgca	agacttctgg	ttactcattc	actggccaca	ccatgaactg	ggtgaagcag	480
agccatggaa	agaaccttga	gtggattgga	cttattaata	cttacaatgg	tgataactaac	540
tacaaccaga	agttcaaggg	caaggccaca	tttactgtag	acaagtcgtc	cagcacagcc	600
tacatggagc	tcctcagtct	gacatctgag	gactctgcag	tctattactg	tgcaaggagg	660
gttacggact	ggtacttcga	tgtctggggc	gcaggggacca	cggtcaccgt	ctcctcagct	720
agcgggtggcg	gtggctccgg	tctagacacc	gtgagcttta	gcactaaagg	tgccacttat	780
attacctacg	tgaatttctt	gaatgagcta	cgagttaaata	tgaaacccga	aggtaacagc	840
catggaatcc	cattgctgcg	caaaaaatgt	gatgatcctg	gaaagtgttt	cgtttttggt	900
gcgctttcaa	atgacaatgg	acagttggcg	gaaatagcta	tagatgttac	aagtgtttat	960
gtggtgggct	atcaagtaag	aaacagatct	tacttcttta	aagatgctcc	agatgctgct	1020
tacgaaggcc	tcttcaaaaa	cacaattaa	acaagacttc	attttggcgg	cagctatccc	1080
tcgctggaag	gtgagaaggc	atatagagag	acaacagact	tgggcattga	accattaagg	1140
attggcatca	agaaacttga	tgaaaatgcg	atagacaatt	ataaaccaac	ggagatagct	1200
agttctctat	tggttggtat	tcaaatggtg	tctgaagcag	ctcgattcac	ctttattgag	1260
aaccaaatga	gaaataactt	tcaacagaga	attcgcccgg	cgaataatac	aatcagcctt	1320
gagaataaat	ggggtaaact	ctcgttccag	atccggacat	caggtgcaaa	tggaatgttt	1380
tcggaggcag	ttgaattgga	acgtgcaaat	ggcaaaaaat	actatgtcac	cgcagttgat	1440
caagtaaaac	ccaaaatagc	actcttgaag	ttcgtcgata	aagatcctaa	agcttaatga	1500

<210> 13

<211> 498

<212> PRT

<213> Artificial

<220>

<223> Polypeptide encoded by the scFv23-gelonin immunotoxin

<400> 13

Met Ser Asp Val Gln Leu Thr Gln Ser Pro Ala Ile Leu Ser Ala

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10

15



Ser	Pro	Gly	Glu	Lys	Val	Thr	Met	Thr	Cys	Arg	Ala	Thr	Pro	Ser	
				20					25					30	
Val	Ser	Tyr	Met	His	Trp	Tyr	Gln	Gln	Lys	Pro	Gly	Ser	Ser	Pro	
				35					40					45	
Lys	Pro	Trp	Ile	Tyr	Thr	Thr	Ser	Asn	Leu	Ala	Ser	Gly	Val	Pro	
				50					55					60	
Ala	Arg	Phe	Ser	Gly	Gly	Gly	Ser	Gly	Thr	Ser	Tyr	Ser	Leu	Thr	
				65					70					75	
Val	Ser	Arg	Val	Glu	Ala	Glu	Asp	Ala	Ala	Thr	Tyr	Tyr	Cys	Gln	
				80					85					90	
Gln	Trp	Ser	Arg	Ser	Pro	Pro	Thr	Phe	Gly	Gly	Gly	Ser	Lys	Leu	
				95					100					105	
Glu	Ile	Lys	Gly	Ser	Thr	Ser	Gly	Ser	Gly	Lys	Ser	Ser	Glu	Gly	
				110					115					120	
Lys	Gly	Val	Gln	Leu	Gln	Glu	Ser	Gly	Pro	Glu	Val	Val	Lys	Pro	
				125					130					135	
Gly	Gly	Ser	Met	Lys	Ile	Ser	Cys	Lys	Thr	Ser	Gly	Tyr	Ser	Phe	
				140					145					150	
Thr	Gly	His	Thr	Met	Asn	Trp	Val	Lys	Gln	Ser	His	Gly	Lys	Asn	
				155					160					165	
Leu	Glu	Trp	Ile	Gly	Leu	Ile	Asn	Pro	Tyr	Asn	Gly	Asp	Thr	Asn	
				170					175					180	
Tyr	Asn	Gln	Lys	Phe	Lys	Gly	Lys	Ala	Thr	Phe	Thr	Val	Asp	Lys	
				185					190					195	

Ser	Ser	Ser	Thr	Ala	Tyr	Met	Glu	Leu	Leu	Ser	Leu	Thr	Ser	Glu	200	205	210
Asp	Ser	Ala	Val	Tyr	Tyr	Cys	Ala	Arg	Arg	Val	Thr	Asp	Trp	Tyr	215	220	225
Phe	Asp	Val	Trp	Gly	Ala	Gly	Thr	Thr	Val	Thr	Val	Ser	Ser	Ala	230	235	240
Ser	Gly	Gly	Gly	Gly	Ser	Gly	Leu	Asp	Thr	Val	Ser	Phe	Ser	Thr	245	250	255
Lys	Gly	Ala	Thr	Tyr	Ile	Thr	Tyr	Val	Asn	Phe	Leu	Asn	Glu	Leu	260	265	270
Arg	Val	Lys	Leu	Lys	Pro	Glu	Gly	Asn	Ser	His	Gly	Ile	Pro	Leu	275	280	285
Leu	Arg	Lys	Lys	Cys	Asp	Asp	Pro	Gly	Lys	Cys	Phe	Val	Leu	Val	290	295	300
Ala	Leu	Ser	Asn	Asp	Asn	Gly	Gln	Leu	Ala	Glu	Ile	Ala	Ile	Asp	305	310	315
Val	Thr	Ser	Val	Tyr	Val	Val	Gly	Tyr	Gln	Val	Arg	Asn	Arg	Ser	320	325	330
Tyr	Phe	Phe	Lys	Asp	Ala	Pro	Asp	Ala	Ala	Tyr	Glu	Gly	Leu	Phe	335	340	345
Lys	Asn	Thr	Ile	Lys	Thr	Arg	Leu	His	Phe	Gly	Gly	Ser	Tyr	Pro	350	355	360
Ser	Leu	Glu	Gly	Glu	Lys	Ala	Tyr	Arg	Glu	Thr	Thr	Asp	Leu	Gly	365	370	375

Ile	Glu	Pro	Leu	Arg	Ile	Gly	Ile	Lys	Lys	Leu	Asp	Glu	Asn	Ala
				380					385					390

Ile	Asp	Asn	Tyr	Lys	Pro	Thr	Glu	Ile	Ala	Ser	Ser	Leu	Leu	Val
				395					400					405

Val	Ile	Gln	Met	Val	Ser	Glu	Ala	Ala	Arg	Phe	Thr	Phe	Ile	Glu
				410					415					420

Asn	Gln	Ile	Arg	Asn	Asn	Phe	Gln	Gln	Arg	Ile	Arg	Pro	Ala	Asn
				425					430					435

Asn	Thr	Ile	Ser	Leu	Glu	Asn	Lys	Trp	Gly	Lys	Leu	Ser	Phe	Gln
				440					445					450

Ile	Arg	Thr	Ser	Gly	Ala	Asn	Gly	Met	Phe	Ser	Glu	Ala	Val	Glu
				455					460					465

Leu	Glu	Arg	Ala	Asn	Gly	Lys	Lys	Tyr	Tyr	Val	Thr	Ala	Val	Asp
				470					475					480

Gln	Val	Lys	Pro	Lys	Ile	Ala	Leu	Leu	Lys	Phe	Val	Asp	Lys	Asp
				485					490					495

Pro Lys Ala

<210> 14

<211> 15

<212> PRT

<213> Artificial Sequence

<220>

<223> 212 Linker (synthetic linker sequence)

<400> 14

Ser Gly Lys Ser Ser Glu G  
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[illegible]